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[54] INHIBITION OF SELECTIN BINDING

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[56] References Cited

U.S. PATENT DOCUMENTS

		Heavner et al
5,486,536	1/1996	Ward et al 514/460
		Rosen et al
		Ratcliffe

FOREIGN PATENT DOCUMENTS

WO 91/19501 12/1991 WIPO . WO 92/07572 5/1992 WIPO . WO 96/34609 11/1996 WIPO .

OTHER PUBLICATIONS

Alon et al., "Lifetime of the P-selectin-carbohydrate bond and its response to tensile force in hydrodynamic flow" *Nature* (1995) 374:539–542.

Arbones et al., "Lymphocyte homing and leukocyte rolling and migration are impaired in L-selectin-deficient mice" *Immunity* (1994) 1:247–260.

Aruffo et al., "CD62/P-selectin recognition of myeloid and tumor cell sulfates" *Cell* (1991) 67:35–44.

Aruffo et al., "Molecular cloning of a CD28 cDNA by high-efficiency COS cell expression system" *Proc. Natl. Acad. Sci. USA* (1987) 84:8573–8577.

Aruffo et al., "Granule membrane protein 140 (GMP140) binds to carcinomas and carcinoma—derived cell lines" *Proc. Natl. Acad. Sci. USA* (1992) 89:2292–2296.

Bertozzi et al., "Sulfated disaccharide inhibitors of L-selectin: Deriving structural leads from a physiological selectin ligand" *Biochemistry* (1995) 34(44):14271–14278.

Brandley et al., "Structure—function studies on selectin carbohydrate ligands. Modifications to fucose, sialic acid and sulphate as a sialic acid replacement" *Glycobiol.* (1993) 3(6):633–641.

Cecconi et al., "Inositol Polyanions" J. Biol. Chem. (1994) 269:15060–15066.

Charych et al., "A 'litmus test' for molecular recognition using artificial membranes" *Chem. Biol.* (1996) 3:113–120. Charych et al., "Direct colorimetric detection of a receptorligand interaction by a polymerized bilayer assembly" *Science* (1993) 261:585–588.

DeFrees et al., "Ligand recognition by E–selectin: Synthesis, inhibitory activity, and conformational analysis of bivalent sialyl lewis x analogs" *J. Am. Chem. Soc.* (1995)117:66–79.

DeFrees et al., "Sialyl lewis x liposomes as a multivalent ligand and inhibitor of E-selectin mediated cellular adhesion" *J. Am. Chem. Soc.* (1996) 118(26):6101–6104.

Dupre et al., "Glycomimetic selection inhibitors: (α–D–Mannopyranosyloxy) Methylbiphenyls" *Bioorg. Med. Chem. Lett.* (1996) 6:569–572.

Foxall et al., "The three members of the selectin receptor family recognize a common carbohydrate epitope, the sialyl lewis" oligosaccharide" *J. Cell. Biol.* (1992) 117(4):895–902.

Frankel et al., "Supramolecular assemblies of diacetylenic aldonamides" *J. Am. Chem. Soc.* (1991) 113:7436–7437.

Fuhrhop et al., "Supramolecular assemblies, a crystal structure, and a polymer of N-diacetylenic gluconamides" *J. Am. Chem. Soc.* (1991) 113:7437–7439.

Geng et al., "Rapid neutrophil adhesion to activated endothelium mediated by GMP-140" *Nature* (1990) 343:757-760.

Hanessian et al., "Design and synthesis of glycomimetic prototypes—a model sialyl lewis* ligand for E-selectin" *J. Syn. Lett.* (1994) 868–870.

Hindsgaul, "Synthesis of carbohydrates for applications in glycobiology" Sem. Chem. Biol. (1991) 2:319–326.

Hiruma et al., "Rational design and synthesis of a 1, 1–linked disaccharide that is 5 times as active as sialyl lewis x in binding to E–selectin" *J. Am. Chem. Soc.* (1996) 118:9265–9270.

Hosomi et al., "Highly stereoselective C-allylation of glycopyranosides with allylsilanes catalyzed by silyl triflate or iodosilane" *Tetrahedron Lett.* (1984) 25:2383–2386.

Hub et al., "Polymerizable phospholipid analogues—New stable biomembrane and cell models" *Angew. Chem. Int. Ed. Engl.* (1980) 19(11):938–940.

(List continued on next page.)

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[57] ABSTRACT

This invention provides compositions for inhibiting the binding between two cells, one expressing P- or L-selectin on the surface and the other expressing the corresponding ligand. A covalently crosslinked lipid composition is prepared having saccharides and acidic group on separate lipids. The composition is then interposed between the cells so as to inhibit binding. Inhibition can be achieved at an effective oligosaccharide concentration as low as 10⁶ fold below that of the free saccharide. Since selectins are involved in recruiting cells to sites of injury, these composition scan be used to palliate certain inflammatory and immunological conditions.

20 Claims, 8 Drawing Sheets